

## Genetic Variation in Response to Salt Stress of Quinoa Grown under Controlled and Field Conditions

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**Abstract**— The objective of this study was to understand the change in response of quinoa genotypes to divers salinity stress conditions e.g in controlled (net-house) and in the different saline fields. The pot experiment was conducted in a net-house at Vietnam National University of Agriculture, Hanoi, Vietnam in spring cropping season to characterize the growth and yield of six quinoa genotypes under four NaCl concentrations (0, 10, 20 and 30 dS m<sup>-1</sup>). At the same time, in Nam Dinh and Hai Phong provinces, two coastal provinces that are most affected by seawater intrusion in the North of Vietnam, same genotypes were studied under two plant densities (20 x 5cm and 50 x 5cm). The results showed that salinity stresses reduced growth and yield characteristics of quinoa plant and varying due to different saline conditions. Plant density of quinoa grown under saline fields was not associated with difference in morphological traits, but might relate to the change in yield characteristics. Salinity stresses reduced plant height, the number of leaves on main stem, the number of branches on plant, head panicle length, dry matter accumulation, 1000-seed weight, individual and grain yield of all quinoa genotypes. However, most of quinoa genotypes produced acceptable yield even under high salt conditions in the field. Among quinoa genotypes, Moradas and Verde adapted well to salt stress conditions with high potential for the number of leaves on main stem, the number of branches on plant, dry matter accumulation and yield than others. These should be recommended varieties for cultivation in saline areas in Vietnam as well as be useful to improve genetic resources in breeding program for salt tolerant quinoa varieties.

**Keywords**— quinoa; salt tolerance; controlled; field; Vietnam

### I. INTRODUCTION

Salinity is the most severe abiotic stress perceived by plants and is affecting 800 million hectares of land worldwide, including 30% of the world's highly productive irrigated land [1]. Salinization is increasing because of poor irrigation management and global climate change. For these reasons, exploiting salt tolerance in crops is an important strategy for plant production development in the near future. Unfortunately, most of food and cash crops such as potato, rice, wheat and maize are "glycophytes" which perform very poor under saline conditions [2]. In addition, breeding for salinity tolerance is difficult as it is controlled by multigenes/QTLs whose expressions are affected by environmental factors [3], [4]. One of important approaches to cope with salinity problems is to directly utilize "halophytes" which are naturally salt tolerant species [5].

Quinoa is a multipurpose nutritious crop, a natural halophyte plant which can be grown in soil conditions with various salinity levels from non-saline soil to extremely saline soil (salt concentration in soil solution is as high as 1/2 salt concentration in the seawater) [5], [6]. No clear seed yield reduction in quinoa grown under highly saline soil

conditions (40 - 50 dS m<sup>-1</sup>) was observed. Interestingly a small seed yield increase was found when quinoa plant grown in saline soil with salinity concentration at rate of 5 - 15 dS m<sup>-1</sup> [7]. Quinoa can grow in high saline soil (350 - 400 mM), whereas yield of other food crops reduced seriously under mild saline condition (40 mM of salinity levels) [2], [8]. Because of good adaptation, quinoa has been produced directly under saline conditions (FAO, 2013) as well as to elucidate the mechanism of its salt tolerance [9]. Quinoa is also known to be more productive under saline conditions than most food crops and considered as a key important crop for the world future food and nutrition security in the context of global climate change [10]. Quinoa seems use several special strategies to acclimate to saline environments and to survive in the soil of salt concentration as high as that in seawater. Therefore, quinoa is an important crop to provide insight understanding of physiology, genetics and molecular of salt tolerance, a complex trait.

Currently, Vietnam has more than one million hectares of land in the coastal areas affected by salinity and prolongable drought. Cultivation soil at these locations is affected by salinity at various levels. In these areas, habitants mainly cultivate conventional crops, such as peanut, maize or

watermelon, etc. However, the yield is very poor for all crops (e.g. less than 1 ton/ha for maize or peanut) and very variable because of high salt concentration in soil. Recently, frequent drought makes cultivation more difficult because of increasing salinization. This study was conducted to understand the genetic variation of quinoa in response to different salt stress conditions e.g controlled versus saline field as well as different salinization in the fields. Six commercial quinoa genotypes introduced from Chile and the Netherlands were characterized for growth and yield under pot experiment where different salt concentrations were added into nutrition solution and irrigated to quinoa growing, at the same time two experiments were conducted in saline fields at Nam Dinh and Hai Phong provinces, these are two locations having severe seawater intrusion that affects crop productions clearly in the North of Vietnam.

## II. MATERIALS AND METHODS

Six quinoa genotypes with different origins, including: three bitter genotypes (saponin presence) Cahuil, Plants Moradas and Plants Verde from Chile; three sweet genotypes (saponin free) Riobamba, Pasto and Atlas from Netherlands were used in this study. Pot and field experiments were conducted as follow:

The pot experiment was conducted under the net-house condition at Faculty of Agronomy, Vietnam National University of Agriculture, Hanoi, Vietnam in spring cropping season, 2015 with four salinity levels: M0- 0 dS m<sup>-1</sup> NaCl (control), M1- 10 dS m<sup>-1</sup> NaCl (mild stress), M2- 20 dS m<sup>-1</sup> NaCl (moderately stress) and M3- 30 dS m<sup>-1</sup> NaCl (extreme stress, comparable to the salt concentration presented in seawater).

In the pot experiment, clean dried sand was mixed with ash of rice straw at 3:1 ratio was used as the plant substrate to fill uniformly in pots 20cm x 20cm. Ten seeds were sown in each pot, after germination young seedlings (2-3 leaves-stage) were thinned and kept 1 seedling/pot. At 5 full leaves-stage, NaCl was added gradually (10 dS m<sup>-1</sup>) until corresponding concentration of each experimental treatments in nutritional solution to irrigate the plant pots for three weeks. The salinity of drainage water and saturated soil extract was monitored to determine the salinity of the substrate, which was adjusted to maintain salinity at predetermined levels [11]. No salt was added to the nutrition solution to use for the control plots and to all pot after three weeks.

At harvest, data were collected for plant height, the number of leaves/main stem and the number of primary branches/plant, and head panicle length of each genotype under different salt levels according method in [12].

Two field experiments were conducted under saline fields in coastal areas (Nghia Hung district - Nam Dinh province and Tran Duong district - Hai Phong province to evaluate growth and yield of 6 quinoa genotypes used in net-house experiment under saline field conditions with two plant densities: M1 (20cm x 5cm as plant density of other conventional crops) and M2 (50 x 5cm as for mechanism cultivation). Salt concentrations in the soil and irrigation water were monitored by sampling three times at sowing day, one month after sowing (flowering stage) and at harvest to

analyse salt concentration by electrical conductivity method [13] and presented in Fig. 1.

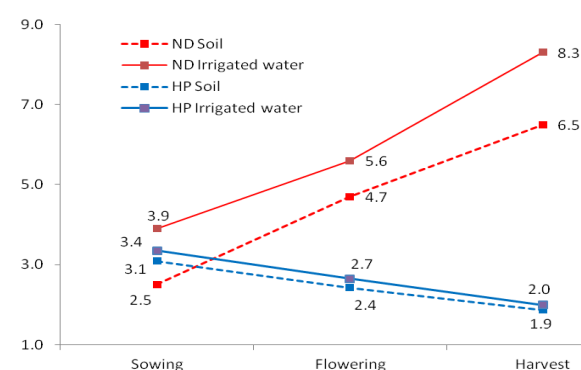


Fig. 1 Salt concentrations in soil and irrigated water at Nam Dinh (ND) and Hai Phong (HP) provinces

In the field experiments, data were collected at 10 days after sowing (DAS), 20 DAS, 30 DAS, 40 DAS, 50 DAS, 60 DAS, and 70 DAS for plant height, the number of leaves/main stem and the number of branches/plant. Dry matter accumulations were determined by constant weight of sampled plant after drying at 80°C in 48 hours at milk stage and harvest time. Growth stages of genotypes were also determined at germination, 2<sup>nd</sup> full leaf, flowering, milk, dough stage and total growth duration from sowing to harvest. At harvest, head panicle length, 1000-seed weight, individual grain yield and yield were determined according methods in [12].

Data analysis, The data were collected and calculated by Microsoft Excel 2010; IRRISTAT 5.0 was used to analysis of variance and calculated Least Significant Different (LSD) at  $p \leq 95\%$ .

## III. RESULTS AND DISCUSSIONS

### A. Pot experiment

The observed that salinity affected growth and yield characteristics of quinoa genotypes with different degrees depending on the salt concentration in the nutrition solutions (Table 1).

The results indicated that salinity levels increasing from 0 dS m<sup>-1</sup> to 30 dS m<sup>-1</sup> reduced plant height, the number of primary branches, the number of leaves on main stem and head panicle length of all quinoa genotypes. Specifically, Cahuil was the best genotype which performed well under normal and salt stress conditions with mean values for plant height, number of primary branches and number of leaves per main stem being 61.7cm, 24.2 branches and 32.0 leaves, respectively. Meanwhile, Atlas genotype was the most affected by salt stress condition for these traits with decreases 3.3cm on plant height, 2.4 branches and 2.8 leaves. However, Cahuil was most salt stress affected genotype for panicle length. Riobamba and Moradas had highest values under saline treatments for panicle length respectively.

TABLE I.  
EFFECT OF SALT STRESSES ON QUINOA GROWN IN POT UNDER NET-HOUSE CONDITIONS

Genotypes	Salt levels	Plant height (cm)	Number of primary branches	Number of leaves/main stem	Head panicle length (cm)
Riobamba	M0	47.7	20.4	31.1	11.4
	M1	47.1	19.6	30.9	11.1
	M2	46.1	19.1	30.4	10.6
	M3	45.5	18.7	29.7	10.0
Atlas	M0	51.1	22.6	33.4	7.0
	M1	50.3	22.1	32.2	6.5
	M2	49.4	21.1	31.6	6.0
	M3	47.8	20.2	30.6	5.7
Pasto	M0	32.5	8.9	20.9	5.3
	M1	32.3	8.6	20.4	5.3
	M2	32.0	8.1	19.9	5.2
	M3	31.8	7.6	19.1	5.0
Moradas	M0	60.0	24.1	30.6	10.4
	M1	59.3	23.6	30.1	10.0
	M2	58.8	22.9	29.3	9.6
	M3	58.0	22.2	28.9	9.1
Cahuil	M0	63.3	25.2	32.8	11.6
	M1	62.0	24.6	32.1	11.0
	M2	61.2	24.1	31.8	10.0
	M3	60.3	22.9	31.2	9.1
Verde	M0	53.9	22.9	29.2	11.3
	M1	53.4	22.1	28.3	10.5
	M2	52.0	21.7	27.9	9.7
	M3	50.8	20.6	27.1	9.0
CV%		2.5	4.7	5.1	11.9
LSD <sub>0.05</sub> (G)		0.88	0.65	1.02	0.72
LSD <sub>0.05</sub> (M)		0.72	0.53	0.84	0.59
LSD <sub>0.05</sub> (G*M)		1.76	1.31	2.05	1.46

## B. Field experiments

There were no significant differences in growing duration, plant height, number of leaves and number of primary branches of quinoa genotypes between two densities (data not shown), therefore data for these traits are showed by average values across two plant densities.

### 1) The time duration in different growing stages of quinoa genotypes

There was no difference in time from sowing to germination among quinoa genotypes, but the differences were found in time from sowing to milk stage, dough stage and especial from sowing to harvest time (Table 2). Pasto and Riobamba genotypes had the shortest total duration (under 85 days), whereas Atlas genotype did the longest with 97 and 107 days at Nam Dinh and Hai Phong provinces, respectively. Atlas also showed the most difference in total duration between two studied locations (10 days), whereas other genotypes showed only 1 to 3 days different.

### 2) Plant height, number of leaves and number of branches of quinoa genotypes

As can be seen from the Figs. 2, 3 and 4, plant height, number of leaves and number of branches of quinoa genotypes increased from sowing to 70 DAS with highest rates during the period from 30 DAS to 60 DAS. All these morphological traits of quinoa genotypes in Hai Phong were higher than those in Nam Dinh which might relate with different salinization regimes of two studied locations (Fig. 1). In fact, at 70 DAS average plant height of genotypes changed from 24.8 to 75.6cm in Hai Phong and from 10.5 to

37.9cm in Nam Dinh provinces. Among genotypes, Verde had the highest plant heights whereas Pasto did the lowest in both locations. In Hai Phong, Verde had the highest number of primary branches, while in Nam Dinh the highest branches number belonged to Modaras. Modaras also had highest leaves number in Nam Dinh (32.3 leaves/stem), and 22.3 leaves/stem, the highest number belonged to Atlas genotype in Hai Phong province. Pasto also showed the lowest values for this trait in both locations.

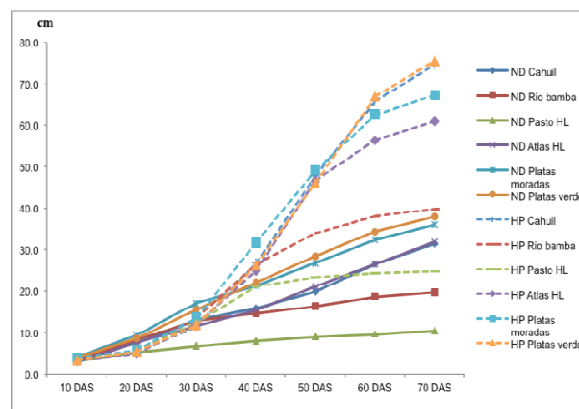


Fig. 2 Plant height of quinoa genotypes at 2 plant densities at Nam Dinh (ND) and Hai Phong (HP) provinces

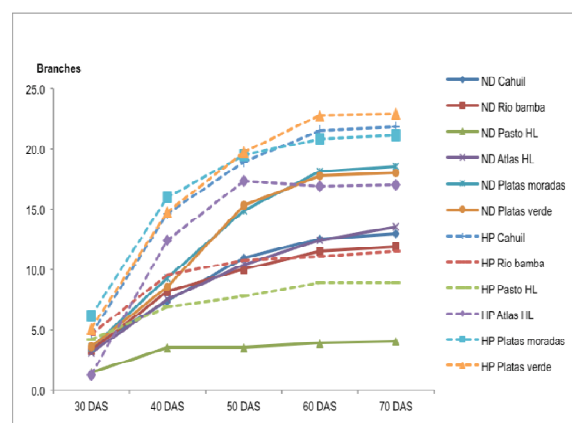


Fig. 3 Number of primary branches of quinoa genotypes at 2 plant densities at Nam Dinh (ND) and Hai Phong (HP) provinces

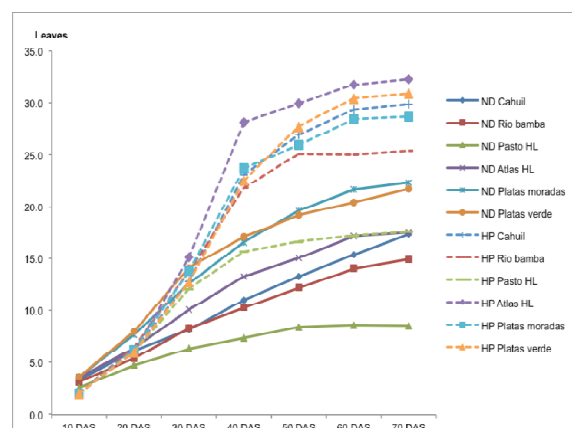


Fig. 4 Number of leaves of quinoa genotypes at 2 plant densities at Nam Dinh (ND) and Hai Phong (HP) province

TABLE II.  
GROWTH DURATIONS OF QUINOA GENOTYPES ACROSS PLANT DENSITIES IN NAM DINH (ND) AND HAI PHONG (HP) PROVINCES

Genotype	Duration from sowing to harvesting (days)											
	Germination		2 <sup>nd</sup> full leaf		Flowering		Milk		Dough		Harvest	
	ND	HP	ND	HP	ND	HP	ND	HP	ND	HP	ND	HP
Cahuil	2	3	10	10	30	36	60	66	78	81	90	93
Riobamba	3	3	12	10	34	34	47	52	61	64	84	85
Pasto	3	3	12	8	27	31	40	45	58	61	79	82
Atlas	3	3	12	11	36	42	65	72	83	85	97	107
Moradas	2	3	10	10	31	36	57	52	75	70	90	91
Verde	3	3	10	9	31	36	60	65	80	82	94	94

3) *Dry matter accumulation of quinoa genotypes in different plant densities:*

Dry matter accumulations (DM) of quinoa genotypes increased from milk stage until to harvest (Table 3). There were significant differences in DM between two plant densities in Hai Phong (where lower density (50cm x 5cm) had higher dry matter accumulation), but not significant in Nam Dinh province. Similar to morphological traits, quinoa genotypes in Hai Phong also had higher DM than those in Nam Dinh province. There were significant differences in DM of studied genotypes. At harvest time among genotypes, Moradas and Verde had the best values for DM and the lowest DM was obtained in Pasto genotype.

TABLE III.  
DRY MATTER ACCUMULATIONS OF QUINOA GENOTYPES AT TWO PLANT DENSITIES IN NAM DINH (ND) AND HAI PHONG (HP) PROVINCES

Plant density	Genotype	Dry matter accumulation at growing stages (g/plant)			
		Milk stage		Harvest	
		ND	HP	ND	HP
M1 (20 x 5cm)	Cahuil	1.71	2.30	8.92	16.10
	Riobamba	1.96	2.23	8.07	6.33
	Pasto	0.74	1.16	2.81	6.00
	Atlas	2.59	2.49	5.90	8.82
	Moradas	2.52	2.61	12.68	16.43
	Verde	2.38	3.01	10.08	19.57
	Mean	1.98	2.30	8.07	12.21
M2 (50 x 5cm)	Cahuil	1.69	2.98	9.88	11.15
	Riobamba	1.94	2.81	7.47	8.05
	Pasto	0.66	1.70	3.21	7.04
	Atlas	2.26	3.12	8.17	9.72
	Moradas	2.32	3.31	10.59	19.02
	Verde	2.29	3.56	11.66	24.27
	Mean	1.86	2.91	8.49	13.21
LSD <sub>0.05M</sub>		0.16	0.06	0.78	0.30
LSD <sub>0.05G</sub>		0.19	0.12	0.57	0.26
LSD <sub>0.05M*G</sub>		0.28	0.17	0.81	0.37
CV%		10.2	4.5	6.8	2.0

4) *Yield and yield components of quinoa genotypes in different plant densities:*

There were significant differences in head panicle length and individual yield, but not significant in 1000-seed weight and grain yield of quinoa genotypes between two plant densities at both experimental locations. The results also showed that yield and yield components of quinoa genotypes in Hai Phong higher than those in Nam Dinh province. Among genotypes, Moradas and Verde had the highest values for all traits, even though Moradas had highest grain yield (1.61 tons/ha) in Nam Dinh, while Verde did the highest (3.64 tons/ha) in Hai Phong province. Pasto genotype had the lowest grain yield with only 1.17 tons/ha and 2.52 tons/ha in Nam Dinh and Hai Phong, respectively.

The results in [14] explained that, salt-induced growth reduction is presumably due to low photosynthetic supply as a consequence of impaired photosynthetic capacity. Also, they confirmed that all growth traits of quinoa plant affected by the very high salinity where, this effect depends on the type and quantity of salt. Our finding showed that under artificial salt stress condition, increasing salt concentrations reduced morphological traits including plant height, number of leaves, number of branches, panicle length (pot experiment). This finding was re-affirmed in the field conditions where salt concentrations in soil and irrigated water in Nam Dinh were much higher than those in Hai Phong (field experiments).

Moreover, although salt concentrations in pot experiment were much higher than field conditions, quinoa genotypes still grew well with lower reductions in all traits when salt levels were increased. Meanwhile, under saline field conditions the reductions were clearer, especially in grain yield of quinoa genotypes. The reasons for this could be that after short term artificial stress for three weeks, quinoa cultivars can recover when rewatering by fresh water, whereas under field conditions plants subject to stress in whole life cycle; and because that while in Hai Phong the stress level increased, in Nam Dinh in opposite trend salt stress level was mollified from sowing to harvest (Fig. 1). It might also suggest that salinity stress at flowering stage might affect quinoa plant more than seeding stage as can be seen from pot versus field experiment.

Our findings are in agreement with [15] that a decreased in number of leaves per plant was found when salt levels increased in irrigated water. Salt concentrations in irrigated water effected on seed germination and early seedling growth of quinoa, where saline stress reduced growth

abilities of quinoa genotypes in comparison with growing in pure water conditions [16]. They also found that morphological properties decreased with increasing the salinity in water. In previous findings, shoot and root weight and total dry matter [11], [16]-[19] decreased under stress conditions in quinoa and others halophyte plant [20], [21]. References [14], [22]-[24] also found the same result in significant reductions in grain yield, number of seeds and seed weight of quinoa in the presence of salinity.

Previous study confirmed that, quinoa plant showed good resistance to water and salt stress through stomatal responses and osmotic adjustments that played a role in the maintenance of a leaf turgor favourable to plant growth and preserved crop yield [25]. Our study found that Moradas and Verde should be potential salt stress tolerant genotypes because of the best performance genotypes for growth and yield characteristics under both artificial and saline field conditions.

TABLE IV.  
YIELD AND YIELD COMPONENTS OF QUINOA GENOTYPES AT TWO PLANT DENSITIES IN NAM DINH (ND) AND HAI PHONG (HP) PROVINCES

Density	Genotype	Head length (cm)		M1000 (g)		Individual yield (g/plant)		Yield (tons/ha)	
		ND	HP	ND	HP	ND	HP	ND	HP
M1 (20 x 5cm)	Cahuil	10.0	9.0	2.54	2.78	2.66	4.25	0.91	2.51
	Riobamba	8.0	8.5	1.61	2.20	2.27	4.77	0.64	2.67
	Pasto	3.3	3.7	-	2.29	-	2.31	-	1.72
	Atlas	7.6	6.5	1.59	2.21	1.67	3.59	0.77	1.82
	Moradas	12.6	8.7	2.49	2.99	4.18	4.97	1.61	3.45
	Verde	17.0	10.2	2.87	3.09	3.65	6.21	1.4	3.64
	Mean	9.7	7.8	1.85	2.59	2.40	4.35	0.88	2.64
M2 (50 x 5cm)	Cahuil	13.4	9.2	2.53	2.83	2.64	6.28	0.64	1.72
	Riobamba	7.3	8.7	1.31	2.31	2.23	4.57	0.55	1.83
	Pasto	3.6	3.8	-	2.33	-	2.59	-	1.05
	Atlas	9.4	6.9	1.42	2.37	1.80	4.45	0.61	1.37
	Moradas	22.3	8.7	2.48	2.99	4.06	5.88	1.17	2.08
	Verde	19.2	10.3	2.82	3.11	3.59	6.21	1.04	2.52
	Mean	12.5	7.9	1.76	2.66	2.38	5.00	0.66	1.76
LSD <sub>0.05M</sub>		0.36	0.94	0.19	0.05	0.33	0.40	0.48	0.13
LSD <sub>0.05G</sub>		0.35	1.54	0.18	0.06	0.42	0.26	0.49	0.17
LSD <sub>0.05M*G</sub>		0.49	2.18	0.26	0.08	0.52	0.36	0.69	0.24
CV%		3.1	3.10	10.1	2.10	3.01	5.10	6.2	7.60

- : No grain harvested

#### IV. CONCLUSIONS

Salinity reduced growth and yield of six genotypes of quinoa under controlled and field conditions. The performance of quinoa under artificial stress for three weeks was different from the fields where salinity stress influences all growing time. Under field conditions, higher saline stress influenced clearly to quinoa growth and yield. It suggests that its worthwhile considering the differences in the responses of quinoa genotypes when studied under artificial salinity stress versus field conditions for future research. Our study also confirmed that at high salt concentration as much as 8 dS m<sup>-1</sup> NaCl most of studied quinoa still produced acceptable yield. Plant density seems having no association with morphological performances of quinoa under saline stress conditions but less populated production might relate with higher yield characteristics of quinoa under saline conditions. Moradas and Verde were the potential salt tolerant genotypes with better growth abilities, higher leaves and primary branches number, dry matter accumulation, 1000-seed weight as well as grain yield in comparison with other genotypes. On the contrary, Pasto showed the lowest

value for all of investigative traits. The contrasting genotypes are recommended for future research to elucidate the mechanisms of salt tolerance in quinoa.

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